PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference ATHCZ/P32969PC	**FOR FURTHER ACTION**	See Form PCT/IPEA/416
International application No. PCT/GB2005/001451	International filing date *(day/month/year)* 15.04.2005	Priority date *(day/month/year)* 15.04.2004
International Patent Classification (IPC) or national classification and IPC INV. A61K38/17 A61K39/395 A61P7/02		
Applicant ATHERA BIOTECHNOLOGIES AB et al.		

1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.
2. This REPORT consists of a total of 13 sheets, including this cover sheet.
3. This report is also accompanied by ANNEXES, comprising:
   a. ☒ *sent to the applicant and to the International Bureau)* a total of 2 sheets, as follows:
      ☒ sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).
      ☐ sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.
   b. ☐ *(sent to the International Bureau only)* a total of (indicate type and number of electronic carrier(s)) , containing a sequence listing and/or tables related thereto, in electronic form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).

4. This report contains indications relating to the following items:

☒	Box No. I	Basis of the report
☒	Box No. II	Priority
☒	Box No. III	Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
☐	Box No. IV	Lack of unity of invention
☒	Box No. V	Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
☐	Box No. VI	Certain documents cited
☐	Box No. VII	Certain defects in the international application
☒	Box No. VIII	Certain observations on the international application

Date of submission of the demand	Date of completion of this report
14.12.2005	31.07.2006
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized officer Böhmerova, E Telephone No. +49 89 2399-7859

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.
PCT/GB2005/001451

Box No. I Basis of the report

1. With regard to the **language**, this report is based on

   ☒ the international application in the language in which it was filed

   ☐ a translation of the international application into , which is the language of a translation furnished for the purposes of:
   - ☐ international search (under Rules 12.3(a) and 23.1(b))
   - ☐ publication of the international application (under Rule 12.4(a))
   - ☐ international preliminary examination (under Rules 55.2(a) and/or 55.3(a))

2. With regard to the **elements*** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report)*:

Description, Pages

1-17	as originally filed

Claims, Numbers

1-6	received on 02.02.2006 with letter of 01.02.2006
7-11	received on 16.06.2006 with letter of 14.06.2006

Drawings, Sheets

1/5-5/5	as originally filed

☐ a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing

3. ☐ The amendments have resulted in the cancellation of:
   - ☐ the description, pages
   - ☐ the claims, Nos.
   - ☐ the drawings, sheets/figs
   - ☐ the sequence listing *(specify)*:
   - ☐ any table(s) related to sequence listing *(specify)*:

4. ☐ This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).
   - ☐ the description, pages
   - ☐ the claims, Nos.
   - ☐ the drawings, sheets/figs
   - ☐ the sequence listing *(specify)*:
   - ☐ any table(s) related to sequence listing *(specify)*:

* *If item 4 applies, some or all of these sheets may be marked "superseded."*

Box No. II Priority

1. ☒ This report has been established as if no priority had been claimed due to the failure to furnish within the prescribed time limit the requested:
   - ☒ copy of the earlier application whose priority has been claimed (Rule 66.7(a)).
   - ☐ translation of the earlier application whose priority has been claimed (Rule 66.7(b)).

2. ☐ This report has been established as if no priority had been claimed due to the fact that the priority claim has been found invalid (Rule 64.1). Thus for the purposes of this report, the international filing date indicated above is considered to be the relevant date.

3. Additional observations, if necessary:

   see separate sheet

Box No. III Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

1. The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non-obvious), or to be industrially applicable have not been examined in respect of:

☐ the entire international application,

☒ claims Nos. 4,5,8,9,10,11

because:

☒ the said international application, or the said claims Nos. 4,5,8,9,10,11 relate to the following subject matter which does not require an international preliminary examination *(specify)*:

see separate sheet

☒ the description, claims or drawings *(indicate particular elements below)* or said claims Nos. 6 (completely), 8-11 (partially) are so unclear that no meaningful opinion could be formed *(specify)*:

see separate sheet

☒ the claims, or said claims Nos. 6 (completely), 8-11 (partially) are so inadequately supported by the description that no meaningful opinion could be formed *(specify)*.

see separate sheet

☐ no international search report has been established for the said claims Nos.

☐ a meaningful opinion could not be formed without the sequence listing; the applicant did not, within the prescribed time limit:

☐ furnish a sequence listing on paper complying with the standard provided for in Annex C of the Administrative Instructions, and such listing was not available to the International Preliminary Examining Authority in a form and manner acceptable to it.

☐ furnish a sequence listing in electronic form complying with the standard provided for in Annex C of the Administrative Instructions, and such listing was not available to the International Preliminary Examining Authority in a form and manner acceptable to it.

☐ pay the required late furnishing fee for the furnishing of a sequence listing in response to an invitation under Rules 13*ter*.1(a) or (b) and 13*ter*.2.

☐ a meaningful opinion could not be formed without the tables related to the sequence listings; the applicant did not, within the prescribed time limit, furnish such tables in electronic form complying with the technical requirements provided for in Annex C-*bis* of the Administrative Instructions, and such tables were not available to the International Preliminary Examining Authority in a form and manner acceptable to it.

☐ the tables related to the nucleotide and/or amino acid sequence listing, if in electronic form only, do not comply with the technical requirements provided for in Annex C-*bis* of the Administrative Instructions.

☐ See separate sheet for further details

Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	1-5,7,9,11
	No:	Claims	8,10
Inventive step (IS)	Yes:	Claims	-
	No:	Claims	1-5,7-11
Industrial applicability (IA)	Yes:	Claims	1-3,6,7
	No:	Claims	-

2. Citations and explanations (Rule 70.7):

see separate sheet

Box No. VIII Certain observations on the international application

The following observations on the clarity of the claims, description, and drawings or on the question whether the claims are fully supported by the description, are made:

see separate sheet

Re Item III

Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

Claims 4,5,8-11 relate to subject-matter considered by this Authority to be covered by the provisions of Rule 67.1(iv) PCT. Consequently, no opinion will be formulated with respect to the industrial applicability of the subject-matter of these claims (Article 34(4)(a)(I) PCT).

Re Item V

Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

Cited documents

Reference is made to the following documents:

D1: US 2003/152513 A1 (BLANKENBERG FRANCIS G ET AL) 14 August 2003

D2: WO 02/067857 A (SURROMED, INC) 6 September 2002

D3: MARI C ET AL: "Annexin V, a new therapeutic tool in atherosclerosis" JOURNAL OF NUCLEAR MEDICINE, vol. 43, no. 5 Supplement, May 2002, page 7P, XP009051427 & 49TH ANNUAL MEETING OF THE SOCIETY OF NUCLEAR MEDICINE; LOS ANGELES, CA, USA; JUNE 15-19, 2002

D4: THIAGARAJAN PERUMAL ET AL: "Inhibition of arterial thrombosis by recombinant annexin V in a rabbit carotid artery injury model" CIRCULATION, [Online] vol. 96, no. 7, 1997, pages 2339-2347, XP002338645

D5: US 2003/170241 A1 (AUKRUST PAL ET AL) 11 September 2003

D6: SHERER Y ET AL: "Immunomodulation for treatment and prevention of atherosclerosis" AUTOIMMUNITY REVIEWS 2002 NETHERLANDS, vol. 1, no. 1-2, 2002, pages 21-27, XP002338646

D7: ALVES J D ET AL: "Atherosclerosis, oxidative stress and auto-antibodies in systemic lupus erythematosus and primary antiphospholipid syndrome" IMMUNOBIOLOGY, FISCHER, STUTTGART, DE, vol. 207, no. 1, 2003, pages 23-28, XP004954257

D8: GOLDENBERG H B ET AL: "Human antibody to phosphorylcholine is bactericidal against Haemophilus influenzae." ABSTRACTS OF THE GENERAL MEETING OF

THE AMERICAN SOCIETY FOR MICROBIOLOGY, vol. 103, 2003, pages D-116, XP009051386 & 103RD AMERICAN SOCIETY FOR MICROBIOLOGY GENERAL MEETING; WASHINGTON, DC, USA; MAY 18-22, 2003

Unless indicated otherwise reference is made to the passages considered relevant in the search report.

The documents D9-D11 were not cited in the international search report.

D9: Database Dissertation Abstract (online) ProQuest Info, Learning; 2002, Binder, Christoph, Johannes: "Defining innate and adaptive immune mechanisms in the atheroprotective effect of immunization with oxidized low-density lipoproteins", Database accession No. AADAA-I3064459

D10: Binder, Cristoph J. ET AL.., Nature Medicine, Vol 9, No. 6, June 2003, pages 736-743

D11: Rose N. ET. AL., Nature Medicine, vol 9, No. 6, 1 June 2003, pages 641-642

Novelty and inventive step

Claims 1-5, 10, 11 (use of annexin V for preventing atherothrombosis and/or plaque rupture; method of treating a subject at risk of atherothrombosis and/or plaque rupture)

The subject-matter of claims 1-5, 10, 11 is considered to be novel in terms of Art. 33(19 and (2) PCT, the reasons being as follows:

Taking into the consideration applicant's arguments put forward in his letter of 14.06.2006, novelty of claims 1-5 over the disclosure of D1 can be acknowledged. D1 relies on the use of annexin V coupled to a radioisotope and/or an effector molecule for detection and treatment of unstable (vulnerable) plaque. It is considered that the complex of annexin V and a radioisotope and/or an effector molecule used in D1 differs from annexin V protein or an N-terminal fragment of annexin V as presently claimed and thus does not fall within the scope of the present claims. D1 further discloses unlabelled annexin V for therapeutic use (see paragraph 31), however this therapeutic use is not precisely defined. The only

disclosure of the therapeutic effect of unlabelled annexin on vulnerable plaque concerns annexin in general or annexin VI (paragraph 33). In conclusion, D1 fails to explicitly disclose the use of unlabelled annexin V for the prevention of atherothrombosis and/or plaque rupture.

D2 teaches the use of modified annexin to prevent thrombosis especially in diabetic patients and during pregnancy and parturition, the prevention of atherothrombosis or plaque rupture is not specifically disclosed.

Although D3 proposes the use of annexin V as a plaque stabilizer in vivo to treat vulnerable plaque, the prevention of atherothrombosis and/or plaque rupture is not explicitly disclosed, thus the subject-matter claimed is novel over D3.

D4 teaches inhibition of arterial thrombosis by annexin V in a rabbit carotid artery injury model. The thrombosis is induced by application of electric current, no effect on atherothrombosis and/or plaque rupture is disclosed.

Claims 1-5, 10, 11 are considered to lack an inventive step under Art. 33(1) and (3) PCT as being obvious over D3 directly proposing annexin V as a suitable plaque stabilizer in vivo to treat vulnerable plaque, which is considered to be identical to the prevention of plaque rupture. This view is supported by the description of the present application stating that the present invention has shown that Annexin V may stabilize atherosclerotic plaque (page 2, lines 15-16). It is considered that the skilled person would have no doubt how to understand the meaning of the phrase 'plaque stabilizer in vivo to treat vulnerable plaque'. Vulnerable plaque, called as well unstable plaque, is known in the art as a plaque with the risk of spontaneous rupture. The meaning of the term 'plaque stabilizer' in the context of vulnerable (i.e. unstable) plaque is considered to be clear and unambiguous. The applicant's argumentation that the meaning of plaque stabilisation according to D3 should differ from that according to the application cannot be followed. Moreover, it cannot be followed how the prevention of macrophage apoptosis shown by D3 should be equal to the increased macrophage proliferation, as alleged by the applicant.

The choice of systemic lupus erythematosus (SLE) patients (claim 5) cannot render the claimed subject matter inventive as atherosclerosis and coronary artery disease are known

in the prior art as the major SLE complications - see D7.

The restriction to the treatment of vulnerable plaques (claim 11) cannot render the subject-matter inventive as D3 specifically mentions vulnerable plaques.

Moreover, the application fails to provide any proof that the claimed solution - administration of annexin V or an N-terminal fragment thereof - actually solves the technical problem posed - provision of an agent to prevent atherothrombosis and/or plaque rupture. The claimed solution appears to be based on the alleged ability of annexin V to stabilize atherosclerotic plaque ("annexin V may stabilize atherosclerotic plaque" -page 2, lines 15-16). However, the stabilizing effect of annexin V is purely hypothetical and is supported neither by experimental data nor by prior art teaching. The application only shows that annexin binds to atherosclerotic plaques, which is however already known from the prior art (see D1), and that the depletion of total IgG from sera with a high capacity to inhibit binding of annexin V restored this binding completely. The connection between the annexin V biding and prevention of plaque rupture is neither directly derivable from the present data nor known from the prior art. As the technical problem has not been solved, no inventiveness can be acknowledged.

Claims 6,8 (use of purified subfraction of pooled immunoglobulin to prevent atherothrombosis and/or plaque rupture; method of treating a subject at risk of atherothrombosis and/or plaque rupture)

The definition of the active agent to be used in claims 6 and 8 - 'purified subfraction of pooled immunoglobulin' is vague and unclear and not sufficiently disclosed, contrary to the requirements of Art. 5 and 6 PCT (see Section VIII). Due to this lack of clarity and disclosure, no comprehensive analysis of novelty and inventiveness of claim 6 and claims 8-11 as far as they concern a method using the purified subfraction of pooled immunoglobulins is practicable at present.

It appears from the description that the subfraction according to claims 6 and 8 may be the one prepared by affinity purification based on binding to a phosphorylcholine conjugate, thus a subfraction comprising anti-phosphorylcholine (aPC) antibodies (see page 7, lines

19-23). Claims 6 and 8 restricted to such subfraction could be considered to be novel over the prior as none of the cited prior art discloses the use of aPC antibodies to prevent atherothrombosis and/or plaque rupture. D9-D11 teach atheroprotective effect of E06/T15 - natural autoantibody to PC - raised endogenously due to the immunization with a pneumococcal vaccine. However, the agents administered in D9-D11 are pneumococcal or MDA-LDL vaccines rather that E06/T15 antibody itself. Neither of D9-D11 teaches or proposes the administration of E06/T15 itself as the active agent to achieve the atheroprotective effect. However, the application fails to prove that such subfraction indeed solves the technical problem posed - the provision of an agent to prevent atherothrombosis and/or plaque rupture. The results on pages 13-14 show that IgG samples that have been depleted (i.e. aPC antibodies removed) allow more annexin V binding than the same antibody sample in which aPC antibodies are present, i.e. that aPC inhibits annexin V binding. If, as proposed by the application, decreased annexin V binding increases the risk of plaque rupture, aPC appear to rise the risk of plaque rupture rather than to prevent it. These results raise further unclarity as to the subfraction to be used according to the present claims.

Claims 7-11 (use of commercially available pooled immunoglobulin for preventing atherothrombosis and/or plaque rupture; a method of treating a subject at risk of atherothrombosis and/or plaque rupture)

The subject-matter of claim 7 appears to be novel over the disclosure of D5 and D6. D5 discloses use of intravenous immunoglobulin (Ivlg) for the treatment of non-viral and non-autoimmune induced heart disorders including coronary syndromes caused by a rupture of an atherosclerotic plaque in one of coronary arteries. D6 discloses immunomodulation including administration of Ivlg for treatment and prevention of atherosclerosis. Ivlg was found to be effective both during fatty streak and plaque formation of atherosclerosis. Neither document discloses the use of Ivlg to prevent atherothrombosis and/or plaque rupture. However, the subject matter of claim 8 is anticipated by the disclosure of D5 and D6 as this claim is directed to a method of treating a subject at risk of atherothrombosis and/or plaque rupture. It is considered that the subjects treated in D5 and D6 falls within the definition of a subject at risk of atherothrombosis and/or plaque rupture as presently claimed. No distinction can be made between the patient to be treated according to the

present claims and those of D5 and D6 as patients at risk of atherothrombosis and/or plaque rupture are prima facie patients having such plaques, i.e. atherosclerotic patients. As D5 discloses treatment of humans, novelty of claim 10 is anticipated by the disclosure thereof.

Claim 9 appears to be new but could not be considered inventive as being obvious in view of at least combination of D6 and D7 for the reasons stated above for claim 5.

Moreover, the claimed solution is not shown to solve the technical problem posed. The examples show only the effect of IgG depleted serum on annexin V binding, no effect of the pooled immunoglobulins on prevention of plaque rupture and/or atherothrombosis is shown. Moreover, the claimed solution appears to encompass embodiments which obviously do not solve the technical problem. Pooled immunoglobulins from patients with SLE are shown to decrease the annexin V binding. Thus in lite of the alleged ability of annexin V binding to stabilise atherosclerotic plaques, pooled immunoglobulins of SLE patients would lead to the destabilisation rather than stabilisation of atherosclerotic plaques. As the technical problem has not been solved and the claimed solution encompass non-working embodiments, no inventiveness can be acknowledged.

Industrial applicability

Subject-matter of claims 1-3, 6, 7 is considered to be industrially applicable under Art. 33(1) and (4) PCT.

For the assessment of the present claims 4,5,8-11 on the question whether they are industrially applicable, no unified criteria exist in the PCT Contracting States. The patentability can also be dependent upon the formulation of the claims. The EPO, for example, does not recognize as industrially applicable the subject-matter of claims to the use of a compound in medical treatment, but may allow, however, claims to a known compound for first use in medical treatment and the use of such a compound for the manufacture of a medicament for a new medical treatment.

Re Item VIII

Certain observations on the international application (clarity)

Claims 6,7 do not meet the requirements of Article 6 PCT in that the matter for which protection is sought is not clearly defined.

Claim 6 attempts to define the subject-matter in terms of the result to be achieved, which merely amounts to a statement of the underlying problem, without providing the technical features necessary for achieving this result. It is directed to the use of a purified subfraction of pooled immunoglobulin defined by the desirable function thereof, namely the ability thereof to inhibit antibodies binding to annexin V or to promote binding of annexin V to endothelium. Moreover, the agent itself is defined by the way of preparation thereof - as a subfraction purified from pooled immunoglobulin, no structural characteristic of the subfraction or fractionating method leading to said subfraction is defined, rendering the subject-matter further unclear. In order to perform the claimed invention, the skilled person would need to isolate all possible subfractions of pooled immunoglobulin and test them for their capacity to inhibit antibodies binding to annexin V, or to promote binding or annexin V to endothelium, which is considered as undue burden.

Although the claims cover almost unrestricted number of possible subfractions of pooled immunoglobulin with the capacity to inhibit antibodies binding to Annexin V or to promote binding of annexin V to endothelium, the application does not provide support for any such serum subfraction. The only serum fraction which appear to be shown to increase binding of annexin to endothelium is the IgG-depleted plasma (see page 13, paragraphs 2, 3). No other subfraction increasing binding of annexin to endothelium is disclosed. No subfraction inhibiting antibodies binding to annexin V is shown by the application. In conclusion, the application does not provide sufficient information allowing a skilled person to perform the claimed invention over the whole scope claimed without undue burden, contrary to the requirements of Art. 5 and 6 PCT.

The term 'commercially available pooled immunoglobulin preparation' employed in claim 7 has no precise meaning as it is not internationally accepted as a standard descriptive term, thereby rendering the definition of the subject-matter of this claim unclear. In this respect, it my not be guaranteed that the product referred to does not have different composition

when purchased from different producers or that the composition does not change with time.

Claim 9 is formulated as directed to a method or a use which renders the scope of protection sought unclear.